

## PATENT APPLICATION TRANSMITTAL LETTER

Docket Number: 8020/002

To the Commissioner of Patents and Trademarks:

Transmitted herewith for filing is the patent application of

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entitled: CHELATED 8-HYDROXYQUINOLINE AND USE HEREOF IN A METHOD OF TREATING EPITHELIAL LESIONS

Enclosed are:

- ☒ 21 pages of written description, claims and abstract.  
☒ 2 sheets of Drawing (2 sets).  
☐ an Assignment of the invention to \_\_\_\_\_  
☒ Declaration of the inventors (unsigned)  
☐ a certified copy of a \_\_\_\_\_ application.  
☐ Associate power of attorney.  
☒ a verified statement to establish small entity status under 37 CFR 1.9 and 1.27 (unsigned)  
☐ information disclosure statement.  
☐ preliminary amendment.  
☐ other: \_\_\_\_\_

## CLAIMS AS FILED

	NUMBER FILED	NUMBER EXTRA	SMALL ENTITY		LARGE ENTITY	
			RATE	FEE	RATE	FEE
BASIC FEE			\$ 395	\$395.00	\$ 790	
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☒ A check in the amount of \$ 538.00 to cover the filing fee is enclosed.☒ The Commissioner is hereby authorized to charge and credit Deposit Account No. 04-1697 as described below. I have enclosed a duplicate copy of this sheet.☐ Charge the amount of \$ \_\_\_\_\_ as filing fee.☒ Credit any overpayment.☒ Charge any additional filing fees required under 37 CFR 1.16 and 1.17.☐ Charge the issue fee set in 37 CFR 1.18 at the mailing of the Notice of Allowance, pursuant to 37 CFR 1.311(b).

Date

February 10, 1998

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## CERTIFICATE OF MAILING (37 CFR 1.10)

I hereby certify that this paper (along with any paper referred to as being attached or enclosed) is being deposited with the United States Postal Service on the date shown below with sufficient postage as Express Mail in an envelope bearing Label No. EL014639819US and addressed to: Box PATENT APPLICATION, Assistant Commissioner for Patents, Washington, DC 20231.

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# CHELATED 8-HYDROXYQUINOLINE AND USE THEREOF IN A METHOD OF TREATING EPITHELIAL LESIONS

## BACKGROUND OF THE INVENTION

### 1. *Field of the Invention*

The present invention pertains to the field of oxinates and, particularly, to the therapeutic use of chelated metal 8-hydroxyquinolines in the treatment of cancers, precancerous lesions, and other abnormal tissues.

### 5 2. *Statement of the Problem*

A preferred treatment modality for many cancers is surgical excision of the cancerous lesion. Surgical excision is not always desirable when surgery could sever nerves or produce scars that interfere with normal movements in tissues proximate the site of surgery.

10 Chemicals have been developed to treat cancers. Rate-sensitive cytotoxic drugs have cytotoxic effects on all tissue types, but are particularly effective against certain cancers that function at a metabolic rate greater than the metabolic rate of normal cells. The increased metabolic rate of these cancers makes them more susceptible to the cytotoxicity of the drugs. In this manner it is possible to provide  
15 a dosage that is fatal to cancer cells, while that same dosage is not fatal to normal cells. By way of example, United States patent 5,684,169 teaches the formation of cyclodextrin complexes of taxol to improve the solubility of taxol in water. Taxol is a cytotoxic drug that is believed to attack and kill cancer cells with a rate-sensitive effect.

20 Other chemicals are specifically designed for the treatment of skin cancers and cancers that reside close to the skin. United States patent 5,605,700 describes a transdermal preparation containing toremifene for use in treating cancers of the skin or cancers that reside a short distance from the skin, such as metastatic lesions of breast cancer. The transdermal preparations are said to be of particular interest  
25 in the treatment of melanoma, lymphoma, Kaposi's sarcoma, and fungoides mycosis. Escharotics or caustic chemicals, such as Podophylin or trichloroacetic acid which are used in the treatment of skin cancer, venereal warts and human papilloma virus, are designed to produce a chemical burn that destroys substantially all of the tissues in contact with the chemical. None of these  
30 chemicals is selective in destroying only the lesion while leaving behind healthy

tissues that exist close to the site of the lesion, i.e., the chemicals lack selectivity and specificity.

The chemical 8-hydroxyquinoline is not used for the treatment of epithelial lesions, such as epitheliomas and venereal warts. 8-hydroxyquinoline is known by various other names including oxine, 8-hydroxy-chinolin, hydroxybenzopyridine, and 8-oxyquiniline. The federal government has approved 8-hydroxyquinoline for cosmetic use in low concentrations of less than five percent. At these concentrations, 8-hydroxyquinoline functions as a cosmetic biocide, as reported in JACT 11(4), 1992. United States patent 4,302,467 reports the use of 8-hydroxyquinoline or its chelates in combination with dehydroacetic acid or sorbic acid. The combination is reported to work synergistically against bacteria and fungus. According to the Merck Index 11th Edition, Merck & Co. publ., p. 4779. (1989), 8-hydroxyquinoline is used as a fungistat, a chelating agent in the determination of trace metals, and a disinfectant.

Chapter ten of the book Albert, Selective Toxicity 3rd Ed., New York, John Wiley & Sons, Inc. (1965) states on page 370-378 that the antibacterial action of oxine is due to chelation. Iron-chelated forms of oxine are toxic to gram-positive bacteria when mixed with  $\text{Fe}^{3+}$  at a 1:1 molar ratio and a 1:2 molar ratio of oxine to iron, but toxicity diminishes at a 1:3 ratio. Despite its strong antibacteriological effects, oxine and its derivatives are not normally injected into the bloodstream because they are inactivated by red blood cells, which secrete a substance that binds oxine.

There remains a need for a topically applied therapeutic composition with selective toxicity in the treatment of epithelial lesions.

## SOLUTION

The present invention overcomes the problems that are outlined above by providing a topically applied therapeutic composition having selective toxicity in the treatment of epithelial lesions. The therapeutic composition demonstrates selective toxicity on human lung cancer, breast cancer, and melanoma. Furthermore, the composition has been used to treat human patients where it demonstrates a one-hundred percent therapeutic index with selective toxicity against venereal warts,

male veruoca warts, lesions produced by human papilloma virus, basal cell carcinoma, solar keratosis, and Kaposi's sarcoma. In veterinary applications where dogs, cats, and horses are the patients, the composition shows a one-hundred percent therapeutic index with selective toxicity against eye cancer, sarcoids, 5 sarcoma, malignant melanoma, rectal adenoma, histiocytoma, and sebaceous adenoma. In this context, the therapeutic index is defined as the number of patients cured divided by the number of patients treated.

A therapeutic composition according to the present invention contains 8-hydroxyquinoline or a functional homologue thereof, a chelatable metal agent, and 10 a carrier. The 8-hydroxyquinoline and the chelatable metal agent are present in effective amounts for treating mammalian epithelial lesions selected from the group consisting of cancerous lesions, precancerous lesions, and warts. These mammalian epithelial lesions specifically include venereal warts, male veruoca warts, lesions produced by human papilloma virus, basal cell carcinoma, solar 15 keratosis, Kaposi's sarcoma, eye cancer, sarcoids, sarcoma, malignant melanoma, rectal adenoma, histiocytoma, and sebaceous adenoma.

Therapeutically effective amounts include therapeutically effective concentrations, as well as therapeutically effective ratios of the 8-hydroxyquinoline and the chelatable metal agent. The preferred range for therapeutically effective 20 ratio is one wherein the molar ratio of 8-hydroxyquinoline to the chelatable metal agent ranges from 1:1 up to 1:N, where N is the oxidation state of the chelatable metal. At the ratio of 1:N, the chelated oxine is easily transported across the cell membrane because it has a neutral charge. Charged species are also observed to be therapeutically effective. Where the metal has a valence of +2, 25 therapeutically effective ratios range from 1:1 to 1:3, and the most preferred ratio is about 1:2. The 8-hydroxyquinoline is preferably present in an amount ranging from five percent to twenty percent of the composition by weight, and the most preferred amount of 8-hydroxyquinoline is about ten percent by weight.

As stated above, the therapeutic compositions include a chelatable metal 30 agent. This agent is preferably a chelatable metal bonded to a halogen, which yields an ionic species of the metal in solution. This reaction is free for covalent and coordinate bonding with oxine in solution. The metal is preferably a heavy

metal or a transition metal. Heavy metals are more preferred, and the preferred heavy metals are copper, iron, manganese, molybdenum, and cobalt. Zinc is the most preferred heavy metal. The metal is preferably provided as a salt including a metal and a halogen. Chlorine is the most preferred halogen. Thus, the most preferred chelatable metal is provided as zinc chloride.

The carrier is preferably formed of a gel base to enhance the time during which the composition is retained on the epithelial lesion where it is applied. Lecithin is preferably added as a penetrating agent, and the penetrating effect of lecithin may be enhanced by the use of dimethyl sulfoxide (DMSO). The carrier also contains an emollient, such as isopropyl palmitate or isopropyl myristate. The gel base is made of a hydrophilic polymer, such as polyacrylamide or polyoxyalkylene derivatives of propylene glycol. A topical steroid, such as 1-2% hydrocortizone may be added to decrease the incidence or severity of contact dermatitis proximate the site of topical application.

A further preferred aspect of the invention is the use of antioxidants to stabilize chelated 8-hydroxyquinoline *in vivo*. Noridhydroguaiaretic acid, derivatives of noridhydroguaiaretic acid, and functional homologues of noridhydroguaiaretic acid stabilize zinc-chelated 8-hydroxyquinoline against inactivation by thermolabile substances in the blood for solutions that are injected into the body. Similarly, 8-hydroxyquinoline and zinc chloride coprecipitated with sodium ascorbate forms a stable orally administrable product having therapeutic efficacy.

The composition is administered to the epithelial lesion by topical application, injection, or oral administration. Where the lesion is scarred or thickening is observed, topical efficacy is enhanced by perforating the lesion with a needle or scalpel prior to topical application of the ointment, in order to enhance the penetration of the ointment into the lesion. Only one topical application is typically required, and the tissue forming the lesion is expected to turn black and necrotic after about three to four weeks. The dead tissue is peeled away, and the wound site typically heals to completion after another three to four weeks have passed. Even where large lesions have been removed in this manner, there is typically no scarring, and even the hair follicles are restored to full function.

## BRIEF DESCRIPTION OF THE DRAWINGS

FIGURE 1 depicts the chelation mechanism between a metal and oxine.

## DETAILED DESCRIPTION OF THE PREFERRED EMBODIMENT

5        FIG. 1 depicts the chelation mechanism for 8-hydroxyquinoline. A chelatable metal M 100 has an oxidation state of  $N^+$ . The chelatable metal M 100 reacts with 8-hydroxyquinoline 104 to produce a 1:1 metal oxinate 104 having a valence of  $N-1$ . A second 8-hydroxyquinoline molecule 106 reacts with the 1:1 metal oxinate 104 to produce a 1:2 metal oxinate 108. This process is repeated  $N$  times until the  
10        resultant oxinate has a neutral charge. even then there is coordinate attraction between the metal M 100 and the heterocyclic nitrogen atoms in other oxinates . The lowest ratio oxinates, e.g., the 1:1 oxinate 104, are the most stable products due to the steric hindrance that the large oxinate ligands provide as the reaction proceeds. Thus, if the reaction is limited by the 8-hydroxyquinoline reagent, the  
15        lower ratio species will predominate. Oxine has a high avidity for metals, and the reaction goes to substantial completion.

      Ionic reaction products, i.e, those having a ratio less than 1: $N$ , are subjected to partition effects where cell membranes regulate the intake of cations. On the other hand, uncharged complexes are liposoluble, and readily penetrate cell  
20        membranes in a manner in which the cell cannot regulate. While 8-hydroxyquinoline is normally not toxic, chelated forms of 8-hydroxyquinoline do have cytotoxic effects in mammalian cells. This cytotoxic effect is empirically observed for both the charged and uncharged reaction products.

      A general theory of pharmacokinetics in the invention is that abnormal  
25        mammalian epithelial cells have membranes which are more permeable to chelated forms of 8-hydroxyquinoline than are normal cell membranes. Thus, the chelated 8-hydroxyquinoline is transported across abnormal cell membranes in a greater concentration than is permitted by normal cell membranes. It follows that topically applied chelated 8-hydroxyquinoline has a greater cytotoxic effect upon the  
30        abnormal cells because abnormal cells absorb it in a greater concentration. This theory accounts for the observed selective toxicity to epidermal lesions.

In addition to topical applications of the compositions, nordihydroguaiaretic acid is used to stabilize the chelated 8-hydroxyquinoline in solutions that are injected into the patient. Nordihydroguaiaretic acid is an antioxidant that protects the chelated oxine compositions against inactivation that, otherwise, might occur due to secretions from the blood that tend to inactivate oxine. Similarly, another antioxidant, sodium ascorbate, is coprecipitated with oxine and zinc chloride from a solution of ethanol and water to form an orally administrable product, which also has *in vivo* utility.

Patients sometimes report a burning sensation at the lesion after topical application of the chelated oxinates. Therefore, it is preferred to place an emollient into the composition, in order to sooth the burning sensation. Penetrating agents, such as lecithin or DMSO, are believed to enhance the efficacy of the compositions.

The following working examples set forth preferred methods and materials for use in practicing the invention.

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#### EXAMPLE 1

##### PREPARATION OF A ZINC-CHELATED LIQUID SOLUTION OF 8-HYDROXYQUINOLINE FOR USE IN TREATING EPITHELIAL LESIONS

Bulk stock solutions were made for later use as intermediates in mixing the final solution. The bulk stock solutions included a gel base solution that was used to stabilize the final product and impart a suitable thickness for the intended environment of use. A lecithin/isopropyl palmitate solution was also prepared.

The gel base solution was prepared by mixing 20 grams (gm) of Pluronic F127<sup>TM1</sup> with 0.2 gm potassium sorbate. Purified water was added to bring the total volume to 100 ml. Pluronic F127<sup>TM1</sup> is a high molecular weight polyoxyalkylene ether derivative of propylene glycol having water soluble, surface active, and wetting properties. The ingredients were mixed in a Braun mixer, sealed in a bowl, and stored under refrigeration at about 5°C once all of the granules became wet. The solution was refrigerated to avoid solidification of the gel, which occurs at room temperature.

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<sup>1</sup> Pluronic F127 is a trademark of the BASF Corporation located in Parisippany, New Jersey.

The lecithin/isopropyl palmitate solution was prepared by dispersing 100 gm of granular soya lecithin and 0.66 gm of sorbic acid in isopropyl palmitate (100 gm or 117 ml). The dispersed mixture was allowed to stand overnight at room temperature of about 20°C. A liquid formed having a syrup-like consistency.

- 5        A 100 gm aliquot of the gel base solution was mixed with 22 ml of the lecithin/isopropyl palmitate solution and 10 ml of distilled water. The mixture was stirred to visual homogeneity. Ten gm of 8-hydroxyquinoline and 20 gm of ZnCl<sub>2</sub> were added to the mixture, which was again stirred to homogeneity.

## EXAMPLE 2

### 10        **TOPICAL APPLICATION OF A ZINC-CHELATED 8-HYDROXYQUINOLINE SOLUTION TO CURE ATHYMIC NUDE MICE OF CANCEROUS LESIONS CORRESPONDING TO EXPLANTED SAMPLES OF HUMAN LUNG CANCER**

- 15        A live human lung cancer cell line Calu-1 or SW-900, as reported by Fogh, J., et al. **59** J. Natl. Cancer Inst. 221-225 (1977), is an epidural lung cancer having an undifferentiated *in vitro* cytopathology. Cell lines of this type are known to grow tumors of corresponding cytopathology when explanted into athymic nude mice, as reported by Fogh and Giovanella eds., *The Nude Mouse in Experimental and Clinical Research*, Academic Press, London, pp 235-266 (1978) (see especially p. 239). Athymic nude mice may be purchased from specially maintained breeding colonies throughout the United States, such as the breeding colony at the University of Nevada in Reno, Nevada. Athymic nude mice are especially desirable to test the efficacy of anticancer compositions because athymic mice lack immune systems. Thus, the mice do not have immune systems to attack explanted cancers in the mice, and the efficacy of anticancer compositions such cancers is primarily due to the compositions alone.
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The test procedures that are outlined below are substantially the same as procedures that have actually been performed on athymic nude mice. The procedures below have been modified slightly to present the most preferred mode of performing the procedures.

- 30        One hundred and fifty athymic nude mice are obtained from a reputable supplier, e.g., the breeding colony at the Worcester Institute, in Worcester, Massachusetts. The mice are split into a treatment test population of fifty mice, a



no treatment test population of fifty mice, and a control population of fifty mice. The populations are maintained as separate groups using conventional procedures for maintaining breeding colonies of athymic nude mice under sterile conditions.

A human epithelial lung cancer cell line, e.g., the Calu-1, SW-900, or MX-1 lung adenocarcinoma line, is cultured in a suitable growth medium, e.g., 50 ml of Eagle's MEM medium (purchased from Life Technologies, Inc., Gaithersburg, MD), which is supplemented with 10% fetal calf serum and 50  $\mu$ g/ml gentamicin from Sigma Chemical Co. of St. Louis, MO. The cells are maintained in a 5% humidified CO<sub>2</sub> atmosphere at 37°C in 75 cm<sup>2</sup> plastic tissue culture flasks, with media passage at 5-7 day intervals.

A sample of approximately five million cancer cells is taken by 20 gauge trocar and extruded into the subcutaneous tissue at the posterior base of the neck on an athymic nude mouse. The procedure is repeated for each member of the treatment and no treatment test populations. Conventional sterile procedures are used while working with the mice to avoid infecting the mice with pathogens other than the cancer cell line. Live tissue from surgical patients having human lung cancer may be substituted for the cultured cell lines.

Approximately one-hundred percent of the mice can be expected to survive the grafting procedure. If proper explantation procedures are used, as will be understood by those who are skilled in those procedures, approximately one-hundred percent of the explanted cancer cells will produce epithelial lesions proximate the site of trocar insertion into the mouse. The explanted cancer is permitted to grow for approximately two to three weeks until a cancerous epithelial lesion of approximately 1.5 to 2 cm in diameter is externally visible.

The untreated test population will typically die within sixty days. Once deaths begin occurring, which is usually within about thirty days, it is preferred to euthanize the entire untreated test population to prevent unnecessary suffering. The deaths typically occur from strangulation due to growth of the cancerous tumor at the base of the neck.

The topical solution that was produced in Example 1 is removed from refrigerated storage and stirred to substantial homogeneity. A cotton swab is dipped into the solution. An athymic nude mouse having a posterior neck lesion

approximately 1.5 to 2 cm in diameter is selected from the treatment test population. A needle is used to perforate the epidermal portion of the lesion at locations spaced about one millimeter apart from one another. A swab is used to apply a coating of solution over the entire lesion and extending at least one-half cm beyond the visible margins of the lesion. This procedure is repeated for each member of the treatment test population. A single topical application of the solution is the sole and exclusive time that the solution is applied to the lesions.

The treatment test population is monitored for nine months after topical application of the solution. Notes are taken as to the progress of cancer in the mice. The lesions appear inflamed on the first day after topical application. Three to five days after topical application, the lesions appear black with white streaks on the surface. Eight to ten days after the topical application, the lesions appear black and necrotic. Fourteen to sixteen days after the topical application, the lesions have sloughed off. Pathologic examination of tissue samples taken from ten individual mice at the interior margins of the sloughed lesion confirms that no cancer is visibly apparent at the margins. In three to four weeks after topical application, the tissue at the former lesion sites has healed to become substantially indistinguishable from normal tissue with no evidence of scarring. The treatment test population exhibits cachexia or a slight emaciation corresponding to an average weight of about 4 to 5 gm less than the control population.

The treatment test population mice and the control population mice are monitored for nine months after topical application of the solution. One-hundred percent of the treatment test population typically survives the procedure with no evidence of cancer after nine months, i.e., the therapeutic index of the solution approximates one hundred percent. This success is contrasted against a one hundred percent mortality in the untreated test population, if the untreated test population has not been euthanized to prevent unwarranted suffering.

### EXAMPLE 3

**TOPICAL APPLICATION OF A ZINC-CHELATED 8-HYDROXYQUINOLINE SOLUTION TO CURE ATHYMIC NUDE MICE OF CANCEROUS LESIONS CORRESPONDING TO EXPLANTED SAMPLES OF OTHER HUMAN CANCERS**

The procedure of Example 2 is repeated for other cancers including human breast cancer, human colon cancer, and human melanoma. It is noted that lung, breast, and colon cancers are classified as malignant epithelial tumors, while melanoma is a malignant nonepithelial tumor. Thus, the study addresses both epithelial and nonepithelial tumors. Specific cell lines that may be used include SW-613 (breast duct adenoca), SW-620 (colon adenoca), and RPMI-7262 (melanoma). See, for a discussion of these cell lines, Fogh, J., et al. 59 J. Natl. Cancer Inst. 221-225 (1977), and Fogh and Giovanella eds., *The Nude Mouse in Experimental and Clinical Research*, Academic Press, London, pp 235-266 (1978) (see especially pp. 238-239).

The therapeutic index is typically one hundred percent effectiveness against all of these explanted cancers in nude mice, with one hundred percent survivorship through treatment, selectivity for the destruction of diseased tissue alone, and no apparent scarring at the former lesion site. The cancers showed no sign of recurrence in the treated test populations after nine months.

#### EXAMPLE 4

##### PREPARATION OF A ZINC-CHELATED 8-HYDROXYQUINOLINE SOLUTION FOR INJECTION INTO ATHYMIC NUDE MICE

These ingredients are weighed out and combined under sterile conditions: nordihydroguaiaretic acid (30 gm), 3% saline in water (30 gm), 8-hydroxyquinoline (10 gm), and ZnCl<sub>2</sub> (20 gm). The ingredients are stirred to substantial homogeneity, and stored under refrigeration at about 5°C.

#### EXAMPLE 5

##### INJECTION OF ZINC-CHELATED 8-HYDROXYQUINOLINE SOLUTION TO CURE ATHYMIC NUDE MICE OF HUMAN CANCER LESIONS

The studies of Examples 2 and 3 are repeated in an identical manner, except the injectable solution of Example 4 is substituted for the topical solution of Example 1. In each case, a one-half ml quantity of the Example 4 solution is injected directly into subcutaneous tissue proximate the lesion.

The therapeutic index of the injected solution is typically one-hundred percent effectiveness against all of the explanted lung, breast, colon, and

melanoma cancers in athymic nude mice, with one hundred percent survivorship through treatment, and no apparent scarring at the former lesion site. The cancers showed no sign of recurrence in the treated test populations after nine months. The therapeutic effects are superior with necrotic effects being observed at the lesion site approximately three days sooner than in the case of the topical ointment.

## EXAMPLE 6

### VARIOUS FORMULATIONS OF CHELATED 8-HYDROXYQUINOLINE SOLUTIONS FOR USE IN THE TREATMENT OF CANCEROUS AND PRECANCEROUS EPITHELIAL LESION

Table 1 below shows various topical product formulations that may be used to dispense therapeutically effective amounts of 8-hydroxyquinoline and zinc chloride. In Table 1 below, "NDGA" represents nordihydroguaiaretic acid. "P20%" represents a solution that contains 20% Pluronic 127 NF<sup>1</sup> by weight, in addition to 0.2% potassium sorbate, and 79.8% distilled water. "P30%" represents a solution that contains 30% Pluronic 127 NF<sup>1</sup> by weight, in addition to 0.2% potassium sorbate, and 69.8% distilled water. "P40%" represents a solution that contains 40% Pluronic 127 NF<sup>1</sup> by weight, in addition to 0.2% potassium sorbate, and 59.8% distilled water. "Lec/l's" refers to a solution that contains 100 gm granular soya lecithin, 100 gm isopropyl palmitate, and 0.66 gm sorbic acid.

The Pluronic 127 NF<sup>1</sup> solutions are preferred due to pharmacactivity that facilitates the transportation of large molecules through epidermal tissues. In this manner, the gels function as adjuvants to improve the efficacy of chelated 8-hydroxyquinoline compositions. A disadvantage of Pluronic 127 NF<sup>1</sup> gels is that they must be made and stored at temperatures less than 40°F because the gels solidify at higher temperatures. Other gel bases may be used including Aqua Base or other polymers dispersed in a suitable solvent for 8-hydroxyquinoline. Lecithin and other penetrants including dimethyl sulfoxide may also be used within prescribed limits to enhance the activity of the chelated oxyquinoline compositions. In Table 1, the term "q.s." is used to show that a sufficient quantity of gel was added

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<sup>1</sup> Pluronic F127 is a trademark of the BASF Corporation located in Parsippany, New Jersey.

to bring the total weight of the combined ingredients to the weight shown in the column, i.e., 100 gm.

continued

TABLE 1

TOPICAL OINTMENT PRODUCT FORMULATIONS

Sample	8-hydroxy-quinoline (gm)	Zinc Chloride (gm)	Distilled Water (ml)	NDGA (gm)	P20% (q.s. gm)	P30% (q.s. gm)	P40% (q.s. gm)	Quercetin (gm)	Lec/s (ml)	Sb <sub>2</sub> S <sub>3</sub> (gm)
A	-	60	30	-	-	-	-	-	-	40
B	10	20	10	-	100	-	-	-	22	-
C	10	20	10	-	-	-	100	-	22	-
D	10	20	10	-	100	-	-	-	20	-
E	10	20	10	10	100	-	-	-	18	-
F	-	10	10	10	100	-	-	10	18	-
G	10	20	10	-	100	-	-	-	15	-
H	10	20	10	-	100	-	-	-	15	-
I	10	20	10	-	-	100	-	-	18	-
J	-	67.5	35	13.75	-	-	-	17.5	-	-

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**EXAMPLE 7**  
**USE OF ZINC-CHELATED 8-HYDROXYQUINOLINE SOLUTIONS**  
**TO TREAT LESIONS IN HUMANS**

A solution corresponding to Sample B from Table 1 in Example 6 was  
5 applied topically to treat cancerous lesions, precancerous lesions, and warts on  
human patients. Treatment included a single topical application of the ointment,  
and was successful in every case. Table 2 below lists the types of lesions that  
were successfully treated, as well as the number of lesions that were successfully  
treated. The total elapsed time from the topical application to sloughing of the  
10 diseased tissue was approximately twelve days in each case. The patients who  
underwent treatment for venereal warts reported much less discomfort than  
normally exists for chemical removal of venereal warts.

**TABLE 2**  
**SUCCESSFUL USE OF CHELATED 8-HYDROXYQUINOLINE IN HUMANS**

5	<b>Lesion Type</b>	<b>Number of Lesions</b>	<b>Location</b>
	Basal cell	2 1 7 2 1 3 1 3 2 1 2 1 2 4	Left side of nose Right ear Right forehead Forehead Left Cheek Scalp Neck Face Hands Head Left mandibular area Arm Left Temple (Unspecified)
	Solar Keratosis	2 1	Head Right forearm
	Human papilloma virus	1 1	Left chest (Unspecified)
	Venereal warts	2 1	Penis (Unspecified)
10	Male veruoca warts	1	(Unspecified)



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<b>Lesion Type</b>	<b>Number of Lesions</b>	<b>Location</b>
Kaposi's sarcoma	1	Arm
	1	Leg
	1	Wrist
	1	Foot

**EXAMPLE 8**

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**USE OF ZINC-CHELATED 8-HYDROXYQUINOLINE SOLUTIONS  
TO TREAT LESIONS IN ANIMALS**

A solution corresponding to sample 73 from Table 1 in Example 6 was applied topically to treat cancerous lesions and precancerous lesions on animals. Treatment was successful in every case, with the exception of one cat that died of sarcoma before the treatment was concluded. Table 3 below lists the types of lesions that were successfully treated and the type of animal on which treatment was successful.

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**TABLE 3****SUCCESSFUL USE OF CHELATED 8-HYDROXYQUINOLINE IN ANIMALS**

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<b>Lesion Type</b>	<b>Type of Animal</b>
Eye cancer	3 Cows
Sarcoid	4 Horses
Sarcoma	1 Dog (leg)
Malignant melanoma	2 Horses
Rectal adenoma	1 Dog
Sebaceous gland adenoma	1 Dog
Histiocytoma	1 Dog
Sebaceous adenoma (two applications)	1 Dog

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15	<b>Lesion Type</b>	<b>Type of Animal</b>
	Sarcoma	2 Cats (one died before end of treatment)

### EXAMPLE 9

#### TITRATION OF ZINC-CHELATED 8-HYDROXYQUINOLINE SOLUTION TO REDUCE ESCHAROTIC EFFECTS WHILE OPTIMIZING THERAPEUTIC EFFECT

Three samples of ointment were prepared corresponding to Sample B from Table 1, except the lecithin/palmitate was not added and the weight percentage amount of zinc chloride comprised 20%, 40%, 55% and 75%, respectively, for the samples. The solutions were each applied to a population of twenty five athymic nude mice with explanted lung cancer in the manner of Example 1. The 55% and 75% samples produced substantially immediate chemical escharotic effects. The 40% sample produced heightened redness proximate the lesion site, and was therapeutically effective. At a 40% concentration, the ointment has the effect of selectively eating away the lesion before the underlying tissue has opportunity to regenerate. The 20% sample was therapeutically effective without burning, and the underlying tissue has a chance to regenerate without the formation of an eschar, i.e., the 20% solution does not tend to leave a hole at the lesion site, rather, no eschar is produced because the necrotic lesion tissue remains in place while the underlying tissue at least partially regenerates.

It is preferred not to use the Pluronic gels at high concentrations of  $\text{ZnCl}_2$  because it has been observed that the gels degrade over time, and it is believed that the gel degradation is due to an altered solution pH.

### EXAMPLE 10

#### USE OF ZINC-CHELATED 8-HYDROXYQUINOLINE ON HUMAN PATIENTS IN A CLINICAL EVALUATION

**Patient 1:** A man entered a clinic in Mexico City to request treatment of a large growth on his right wrist. The growth was approximately eleven centimeters in diameter and circumscribed more than fifty percent of the wrist. The size and location of the growth meant that surgery would leave the man with impaired wrist

movement. The growth was diagnosed as basal cell carcinoma. A solution corresponding to Sample B of Table 1 was applied to the carcinoma using a cotton swab. The patient was monitored for two months. The patient complained of a burning sensation. The growth appeared red and inflamed at its margins on the first day after treatment. One week later, the growth appeared black and necrotic. Two weeks after treatment, the growth was black and necrotic. The dead growth was peeled away using forceps. pathologic examination of tissue from the wrist confirmed that basal cell carcinoma was absent from healthy tissues at the margin of the wound. Six weeks after treatment, the wound site appeared as healthy tissue with no sign of basal cell carcinoma or scarring. Hair follicles were intact at the site of the former wound and basal cell carcinoma. The man retained full function in the affected wrist.

**Patient 2:** A woman entered a clinic in Mexico City with a basal cell carcinoma approximately on inch below the corner of her left eye. The growth was approximately one-half inch in diameter. The surgical prognosis was that removal of the growth would leave the woman with partial paralysis of the face and eye muscles. A solution corresponding to solution #73 was applied to the carcinoma using a cotton swab. The patient was monitored for two months. The patient complained of a burning sensation. The growth appeared red and inflamed at its margins on the first day after treatment. One week later, the growth appeared black and necrotic. Two weeks after treatment, the growth was black and necrotic. The dead growth was peeled away using forceps. pathologic examination of tissue from the wrist confirmed that basal cell carcinoma was absent from healthy tissues at the margin of the wound. Six weeks after treatment, the wound site appeared as healthy tissue with no sign of basal cell carcinoma or scarring. Hair follicles were intact at the site of the former wound and basal cell carcinoma. The woman retained full function in muscles of the face and eye.

## EXAMPLE 11

### PREPARATION OF AN ORALLY ADMINISTRABLE COMPOSITION

A solution was mixed to include equal 33 gram weights of 8-hydroxyquinoline, sodium ascorbate, and zinc chloride in 90% ethanol with

sufficient water added to just dissolve the ingredients at 110°F. The mixture stood to ambient room temperature under a ventilation hood for twenty four hours. A vacuum aspirator was used to remove remaining liquid while coprecipitating the remaining moieties from solution. The precipitate was scraped from the beaker, 5 supplemented with 5 grams of ascorbic acid, and ground to homogeneity with mortar and pestle.

Those skilled in the art will understand that the preferred embodiments described above may be subjected to apparent modifications without departing from the true scope and spirit of the invention. The inventors, accordingly, hereby state their intention to rely upon the Doctrine of Equivalents, in order to protect their full rights in the invention.

## CLAIMS:

1. A composition for use in treating epithelial lesions comprising:  
8-hydroxyquinoline;  
a chelatable metal agent; and  
a carrier,
- 5 wherein said 8-hydroxyquinoline and said chelatable metal agent are present in effective amounts for treating mammalian lesions selected from the group consisting of cancerous lesions, precancerous lesions, and warts.
2. The composition as set forth in claim 1 wherein said effective amounts include a ratio of 8-hydroxyquinoline to said chelatable metal agent ranging from 1:1 to 1:3 by weight.
3. The composition as set forth in claim 2 wherein said ratio is about 1:2.
4. The composition as set forth in claim 1 wherein said effective amounts are therapeutically effective without producing an eschar.
5. The composition as set forth in claim 4 wherein said effective amounts include zinc chloride as said chelatable metal agent present in an amount ranging from five percent to twenty percent of said composition by weight.
6. The composition as set forth in claim 1 wherein said effective amounts include zinc chloride as said chelatable metal agent present in an amount ranging from five percent to twenty percent of said composition by weight.
7. The composition as set forth in claim 1 wherein said effective amounts are therapeutically effective with producing an eschar.
8. The composition as set forth in claim 7 wherein said effective amounts include zinc chloride as said chelatable metal agent present in an amount ranging up to forty percent of said composition by weight.
9. The composition as set forth in claim 1 wherein said effective amounts include zinc chloride as said chelatable metal agent present in an amount ranging up to forty percent of said composition by weight.
10. The composition as set forth in claim 1 wherein said chelatable metal agent is zinc chloride.
11. The composition as set forth in claim 1 wherein said chelatable metal agent includes zinc.

12. The composition as set forth in claim 1 chelatable metal agent includes chlorine.

13. The composition as set forth in claim 1 wherein said chelatable metal agent is zinc chloride.

14. The composition as set forth in claim 1 wherein said carrier is a gel base.

15. The composition as set forth in claim 1 wherein said gel base is a polyoxyalkylene ether derivative of propylene glycol.

16. The composition as set forth in claim 1 wherein said carrier contains a penetrant.

17. The composition as set forth in claim 1 wherein said penetrant is lecithin.

18. The composition as set forth in claim 1 wherein said penetrant is dimethyl sulfoxide.

19. The composition as set forth in claim 1 wherein said carrier contains an antioxidant.

20. The composition as set forth in claim 1 wherein said antioxidant is selected from a group consisting of nordihydroguaiaretic acid, nordihydroguaiaretic acid derivatives, and functional homologues of nordihydroguaiaretic acid.

21. The composition as set forth in claim 1 wherein said antioxidant is selected from a group consisting of ascorbic acid, ascorbic acid derivatives, and functional homologues of ascorbic acid.

22. The composition as set forth in claim 1 wherein said 8-hydroxyquinoline is derived from quercetin in said composition.

23. A method of treating mammalian epithelial lesions, said method comprising the steps of:

identifying a mammalian lesion selected from the group consisting of precancerous lesions, cancerous lesions, and warts;

- 5 administering to said lesion a composition containing
  - 8-hydroxyquinoline,
  - a chelatable metal agent, and
  - a carrier,

10            wherein said 8-hydroxyquinoline and said chelatable metal agent are present in effective amounts for treating mammalian epithelial lesions selected from the group consisting of cancerous lesions, precancerous lesions, and warts; and  
             permitting said composition to destroy said lesion.

24.    The method of claim 23 wherein said step of administering to said lesion includes a step of providing said composition to include said effective amounts as a ratio of 8-hydroxyquinoline to said chelatable metal agent ranging from 1:1 to 1:3.

25.    The method of claim 24 wherein said ratio is about 1:2.

26.    The method of claim 23 wherein said effective amounts include said 8-hydroxyquinoline present in an amount ranging from five percent to twenty percent of said composition by weight.

27.    The method as set forth in claim 23 wherein said chelatable metal agent is zinc chloride.

28.    The method as set forth in claim 23 further including a step of permitting skin at said lesion site to heal without scarring.

29.    The method as set forth in claim 23 wherein said steps of identifying, administering, and permitting are performed without the formation of an eschar at the lesion site.

30.    The method as set forth in claim 23 wherein said steps of identifying, administering, and permitting are performed with the formation of an eschar at the lesion site.

31.    The method as set forth in claim 23 wherein said step of administering is performed topically.

32.    The method as set forth in claim 23 wherein said step of administering is performed by injection.

33.    The method as set forth in claim 23 wherein said step of administering is performed orally.

**ABSTRACT**

Oximates including 8-hydroxyquinoline and a heavy metal are topically applied to epidermal lesions for therapeutic effect. The zinc oxinate compositions are shown to be therapeutically effective against The therapeutic composition demonstrates selective toxicity with a therapeutic index of one-hundred percent on human lung cancer, breast cancer, melanoma, venereal warts, male veruoca warts, lesions produced by human papilloma virus, basal cell carcinoma, solar keratosis, and Kaposi's sarcoma. In veterinary applications where dogs, cats, and horses are the patients, the composition shows a one-hundred percent therapeutic index with selective toxicity against eye cancer, sarcoids, sarcoma, malignant melanoma, rectal adenoma, histiocytoma, and sebaceous adenoma.



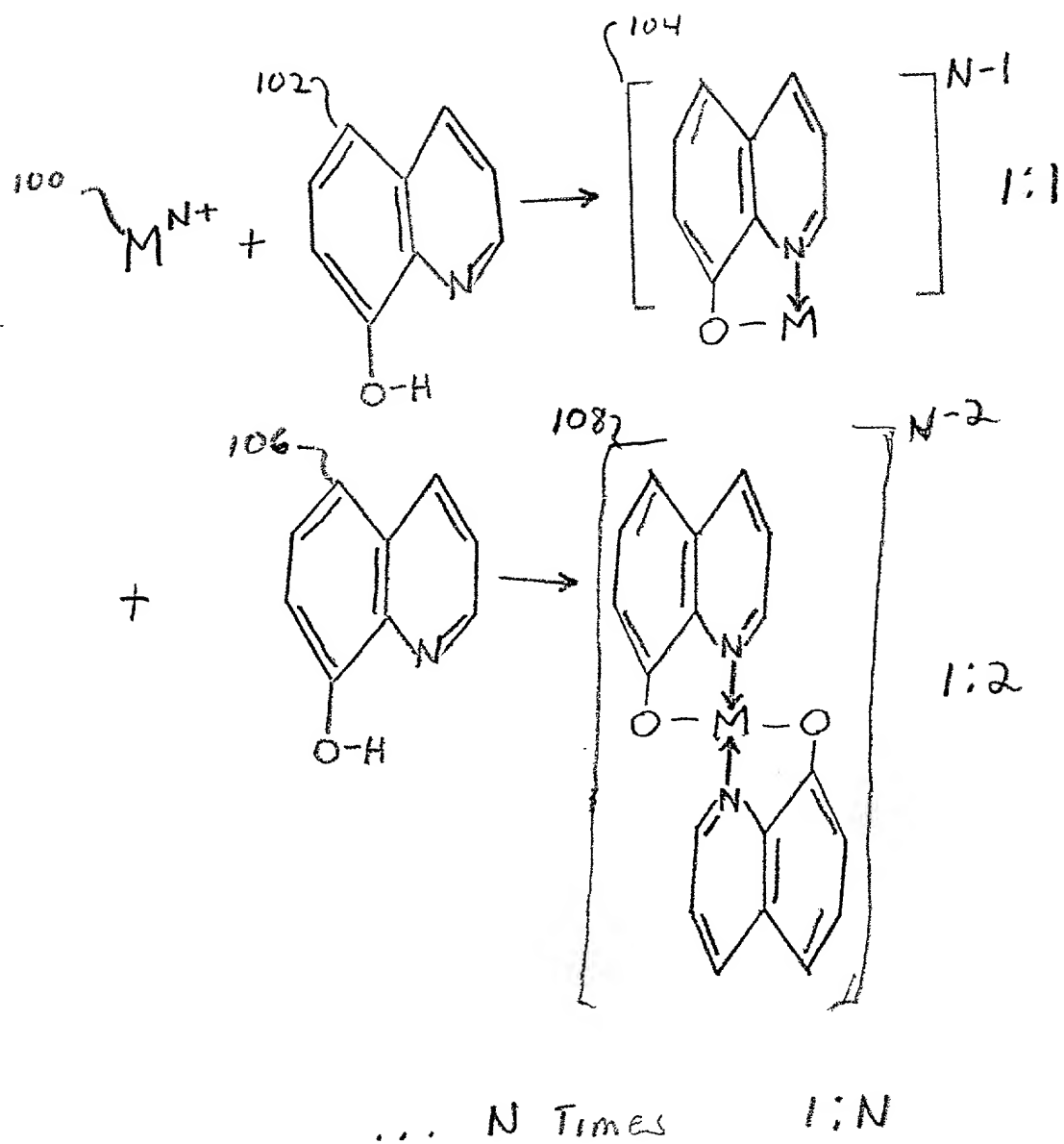


FIG. 1

[illegible]

As a below named inventor, I hereby declare that:

the specification of which is attached hereto unless the following is checked:

I hereby claim foreign priority benefits under Title 35, United States Code, §119 of any foreign application(s) for patent or inventor's certificate listed below and have also identified below any foreign application for patent or inventor's certificate having a filing date before that of the application on which priority is claimed.

Yes      No

(Status - patented, pending, abandoned)

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# DECLARATION FOR PATENT APPLICATION

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Docket Number: 8020/002

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(2/92 PTO)

**VERIFIED STATEMENT CLAIMING SMALL ENTITY STATUS  
(37 CFR 1.9(f) & 1.27(c)) -- SMALL BUSINESS CONCERN**

Docket Number: 8020/002

Applicant or Patentee: Jordan, M.D., et al.

Serial or Patent Number: Not yet known

Filed or Issued: Herewith

Title: CHELATED 8-HYDROXYQUINOLINE AND USE THEROF IN A METHOD OF TREATING  
EPITHELIAL LESIONS

I hereby declare that I am

     the owner of the small business concern identified below:

  X   an official of the small business concern empowered to act on behalf of the concern identified below:

NAME OF SMALL BUSINESS CONCERN Dermex Pharmaceuticals, L.L.C.

ADDRESS 7374 Inspiration Drive, Parker, Colorado 80138

I hereby declare that the above-identified small business concern qualifies as a small business concern as defined in 13 CFR 121.12, and reproduced in 37 CFR 1.9(d), for purposes of paying reduced fees to the United States Patent and Trademark Office, in that the number of employees of the concern, including those of its affiliates, does not exceed 500 persons. For purposes of this statement, (1) the number of employees of the business concern is the average over the previous fiscal year of the concern of the persons employed on a full-time, part-time or temporary basis during each of the pay periods of the fiscal year, and (2) concerns are affiliates of each other when either, directly or indirectly, one concern controls or has the power to control the other, or a third party or parties controls or has the power to control both.

I hereby declare that the rights under the contract or law have been conveyed to and remain with the small business concern identified above with regard to the invention described in:

     the specification filed herewith with title as listed above.

  X   the application identified above.

     the patent identified above.

If the rights held by the above-identified small business concern are not exclusive, each individual, concern or organization having rights in the invention must file separate verified statements averring to their status as small entities, and no rights to the invention are held by any person, other than the inventor, who would not qualify as an independent inventor under 37 CFR 1.9(c) if that person made the invention, or by any concern which would not qualify as a small business concern under 37 CFR 1.9(d), or a nonprofit organization under 37 CFR 1.9(e).

Each person, concern or organization having any rights in the invention is listed below:

  X   No such person, concern or organization exists.

     Each such person, concern or organization is listed below:

FULL NAME: \_\_\_\_\_

ADDRESS: \_\_\_\_\_

[ ] INDIVIDUAL [ ] SMALL BUS. CONCERN PROFIT ORGANIZATION

Separate verified statements are required from each named person, concern or organization having rights to the invention averring to their status as small entities. (37 CFR 1.27)

I acknowledge the duty to file, in this application or patent, notification of any change in status resulting in loss of entitlement to small entity status prior to paying, or at the time of paying, the earliest of the issue fee or any maintenance fee due after the date on which status as a small entity is no longer appropriate. (37 CFR 1.28(b)).

I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under section 1001 of Title 18 of the United States Code, and that such willful false statements may jeopardize the validity of the application, any patent issuing thereon, or any patent to which this verified statement is directed.

NAME OF PERSON SIGNING Frank S. Potestio, M.D.

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SIGNATURE \_\_\_\_\_ DATE \_\_\_\_\_